

LUDWG INSTITUTE E3400

03/04



LUD 5664 (10017134)

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant(s) : Laure DUMOUTIER et al.
Serial Number : 09/626,617
Filing Date : July 27, 2000
For : ISOLATED NUCLEIC ACID MOLECULES WHICH
ENCODE T CELL INDUCIBLE FACTORS, OR
INTERLEUKIN-21, THE PROTEINS ENCODED, AND
USES THEREOF
Art Unit : 1644
Examiner : Phillip GAMBEL

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MAR 03 2004

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION

SIR:

THE UNDERSIGNED hereby declare as follows:

1. We are the properly named, co-inventors of the subject matter set forth in the above-referenced application.
2. We reduced the invention described in the above-referenced application to practice prior to May 27, 1999. We did the work described herein in Belgium, which is a member of the World Trade Organization. The United States is also a member of the World Trade Organization.
3. Attached hereto are copies of relevant pages from our notebooks, evidencing successful showing the stimulation of STAT-3 by both murine and human IL-21/IL-21.

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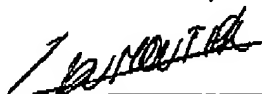
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LUD 5664 (10017134)

4. With respect to the claimed subject matter, this was reduced to practice no later than May 27, 1999.
5. The first set of experiments used murine IL-21/TIF and measured ~~STAT binding to~~ ^{STAT binding to} ~~GRF~~ ^{GRR}. Cell line MES13 is described, for example, in example 21 of the specification, and was a cell line tested, as will be seen in the figures accompanying the experimental details. Indeed, example 21 describes the work in the first part of the attachments. The second set of experiments reached the same conclusions, using human IL-TIF/TL-21.
6. We hereby declare that all statements made herein of our own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

February 17, 2004
Date


Laure DUMOUTIER

February 17, 2004
Date


Jean-Christophe RENAULD

Symbol Report: IL22

[Back To Search](#)

Approved Gene Symbol: IL22
Approved Gene Name: interleukin 22
HGNC ID: 14900
Location: 12q15
Sequence Accession ID: AF279437
PMIDs: [10954742](#), [10875937](#)
Status: Approved

Synonyms

Aliases: ILTIF, IL-21, zcyto18, IL-TIF, IL-D110, TIFa, TIFIL-23, IL-22
Previously Approved Symbols:
Previous Gene Names:

Other Database Links

- **Ensembl:** [IL22](#)
 - **Enzyme ID:**
 - **GENATLAS:** [IL22](#)
 - **GeneCards:** [IL22](#)
 - **GeneClinics/GeneTests:** [IL22](#)
 - **IMGT:**
 - **LocusLink ID:** [50616](#)
 - **MGI:**
 - **OMIM:** [605330](#)
 - **RefSeq ID:** [NM_020525](#)
 - **Swiss-Prot ID:** [Q9GZX6](#)
-

Maintained by nome@galton.ucl.ac.uk Script last updated: July 2002 [HGNC Homepage](#)

EMSA au 17E13 et au BW.

protocole voir LD 323.

Oligos GARS marqué d° LD 323.

preparé des ech.

• extrait nucléaire 4ul (mél. BW / ou IL9).

reprise des extraits nucléaires.

YES 13 - Vide

$10 \times 10^6 \phi$ / 15ml + 250ul
de SN 293
LD 249.

- TIF ens

- TIF Antisens

BW - Vide

- TIF ens

- TIF Antisens.

→ 10' à 37°C

→ + 5ml de PBS froid

→ 10 des ϕ .

→ reprendre d° 1ml de B1.

→ 1.5' à 4°C

→ 1.65ul de NP-40. → Vortexer 10"

→ 10 30"

→ écouler le SN - reprendre le culot
d° 100ul de B2.

→ 30' à 4°C sous agitation

→ reprendre le SN.

extrait nucléaire 4ul

Tp sharp 4ul

Poly dIdc 2ul

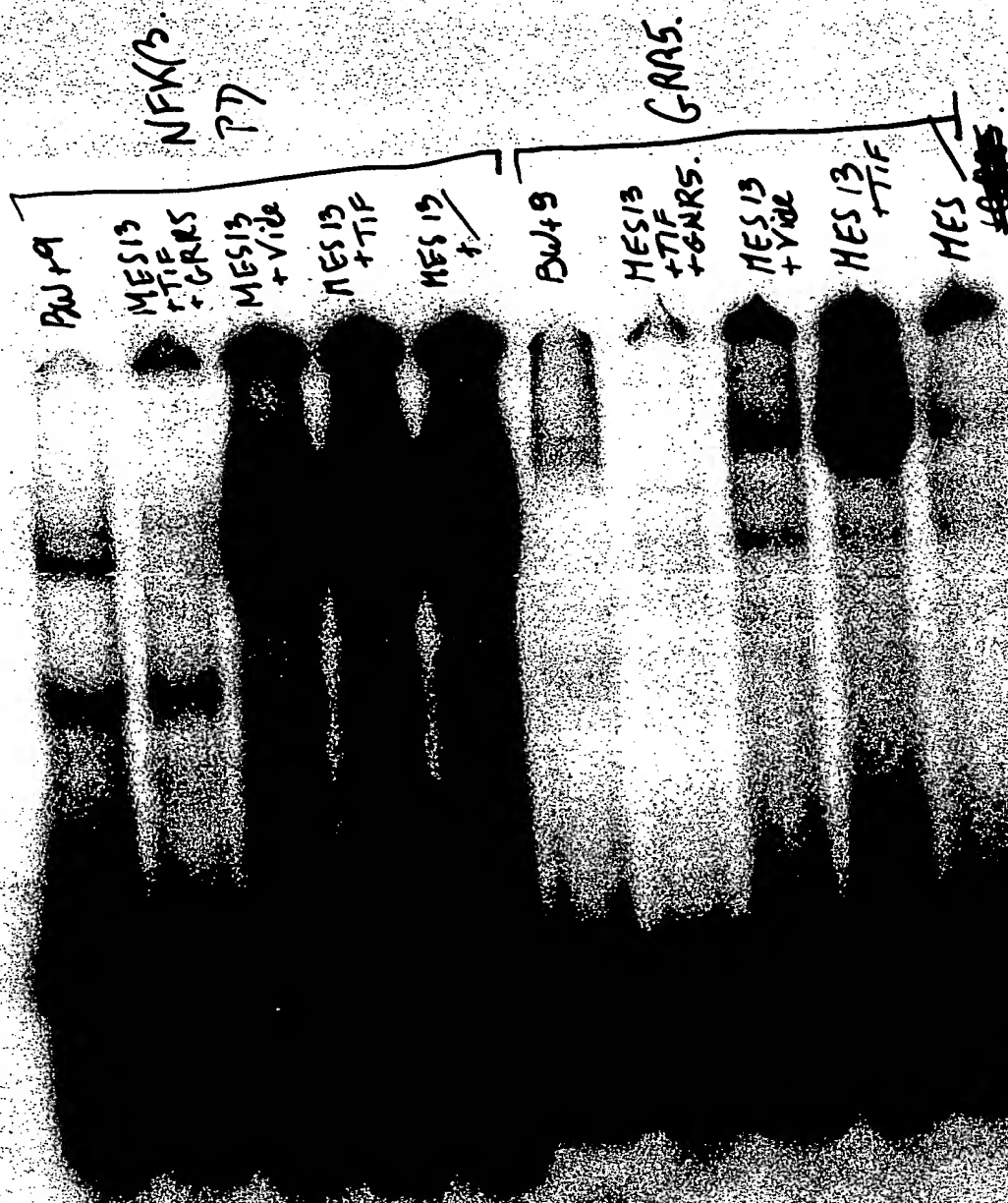
H₂O au complet 9ul

→ 5' a TA.

+ 100 000 Upm.

→ 85' a TA

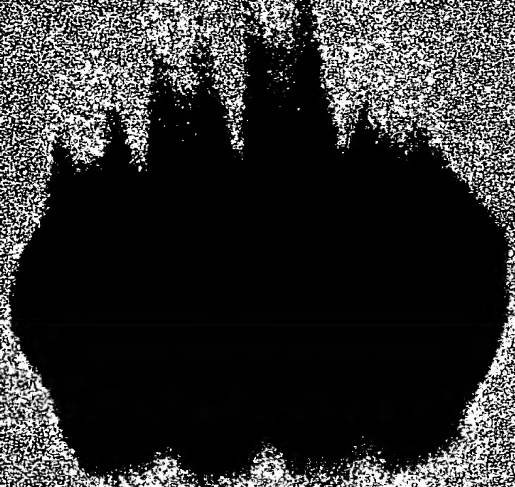
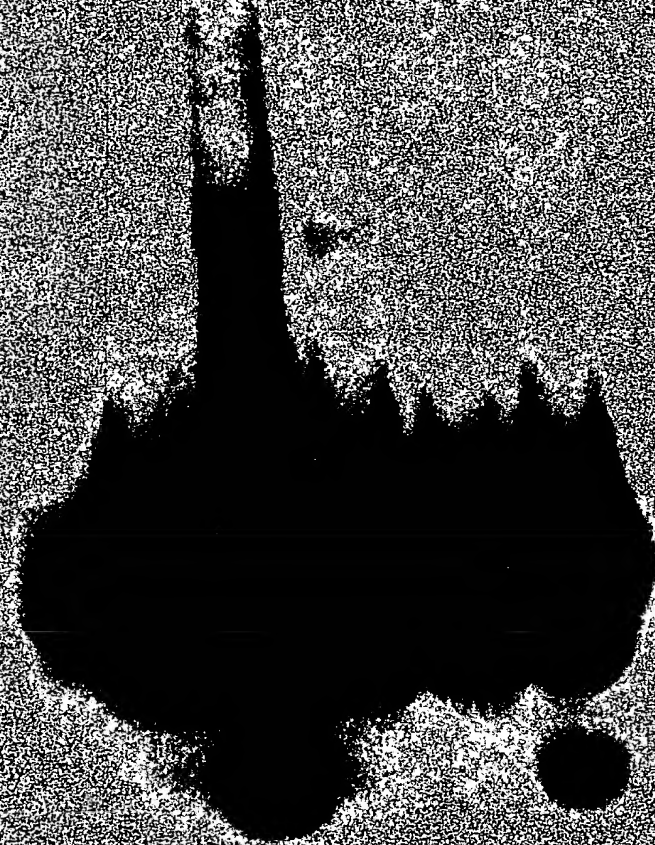
→ de tot sui gel.



Expt over 15
3h.

BW
BW/ Vld
T/F
T/F + AS
T/F + GART
BW/ + DGS

MES 13
Vld
T/F
Sens
A. VAS
Sens + GART



EMSA MES13 stimulé avec htif

LD525

1. marquer l'oligo

5 μ l d'oligo GRR Long.
2,6 μ l de T₄ pol.
1 μ l d'ATP
2,4 μ l d'³²P
5 μ l de H₂O

→ 30' à 37°C

→ chromatographie

→ Comptes μ l = 250.000 cpm
LD à diluer 2,5x

2. préparation des Extraits

→ Comptes les d 33000 d/ml → 8 ml

reprendre d 7 ml
stimuler → 6 conditions :

1 μ l	TIF	SNS	(SN 292)
1 μ l	"	AS	(LD 240)
2,5 μ l	1/2 TIF	1/2 AS	(SN 203)
	1/2 TIF	1/2 AS	(LD 523)
	1/2 TIF	1/2 AS	
	1/2 TIF	1/2 AS	

→ 15' à 37°C

+ 5 ml de P/D S

→ 5' à 1.200 rpm

reprendre le culot d'2 T₄ pol B1 (voir LD 433)

→ 15' à 4°C.

+ 32,5 μ l de NP-40 → le d reprendre
le culot d'2 50 μ l de T₄ pol B2 (voir LD 433)

→ 30' à 4°C sur la roue

→ le et garder le SN

3. préparat° des ech.

extrait nucléaire 4 μ l

tr_{sharp} 5x 4 μ l

polydIdc 4 μ l

→ H₂O 9 μ l a 7 μ l si 2 μ l > STAT3

5' à TA
+ 1 μ l d'oligo à 100 000 cpm/ μ l

85' à TA

charger le gel A + de Tips fin pour charger.

=> j'ai utilisé des Tips vert qui
n'allaient pas à fond du puit

4. gel : Tpon 5ml.

Acrytomide 12,5ml } dégozer.

H₂O 32,5ml

+ 30ul de persulfate 10%.

+ 25ul de TEMED.

→ couler le gel.

→ polymériser 1h.

5. prérun 20ml 2h.

6. run 30ml 1h30

7. écher le gel 1h

8. mettre en autoradio 2h ou O/N.

expo
on
over
TS.



P10 MAX FILM

KODAK BIO MAX FILM

KODAK BIO MAX FILM